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Assessment of Air Quality of Livestock Farms in District Kasur and Lahore

Muhammad Jamil[1], Safdar Sidra[1]*, Ali Hussain[2], Muhammad Imran[3] and Ali Ahmad Sheikh[4]

[1]*Department of Wildlife and Ecology, University of Veterinary and Animal Sciences, Lahore, Pakistan*
[2]*Institute of Zoology, University of the Punjab, Lahore, Pakistan*
[3]*Institute of Biochemistry and Biotechnology, University of Veterinary and Animal Sciences, Lahore, Pakistan*
[4]*Institute of Microbiology, University of Veterinary and Animal Sciences, Lahore, Pakistan*

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ABSTRACT

Agriculture, particularly livestock is the largest sector of Pakistan's economy. While there are on-going efforts for further improvements in this sector, there is dire need to understand the environmental factors involved in well-being of the animals. The current study was conducted to evaluate the air quality of ten livestock farms of Districts Kasur and Lahore in an attempt to document the factors responsible for a better healthy environment for the valuable animals. The farms were selected and categorized on the basis of their respiratory disease outbreaks in the recent past. Particulate matter (PM_{10} and $PM_{2.5}$), bioaerosols, potentially toxic trace elements (PTEs), CO_2, temperature and relative humidity were measured at each farm during the summer and winter season. While the mean PM levels (3.29 ± 1.82 µg/m^3 healthy farms and 3.81 ± 3.67 µg/m^3 for the diseased farms) were below the threshold levels, the various activities around the farms and impact of seasons was significant. The air-borne micro-flora of animal farms comprised of opportunistic pathogens like *Staphylococcus aureus* and *Pasteurella multocida* . To conclude the air of the monitored farm was not so polluted probably due to the natural ventilation that might have played a role in timely dispersal of the air-borne contaminants. However, keeping in view the recurrent disease episodes at some of these farms, it is recommended to regularly monitor the air composition for extended periods so as to identify the factors responsible.

INTRODUCTION

Livestock is an important constituent of the agriculture sector and the most dynamic part of national economy. Livestock contributed about 61.9% towards the agriculture sector in Pakistan and 14% towards the GDP in the year 2021-22 with a growth of 3.76 percent as compared to 2.99 percent last year (Pakistan Economic Survey, 2021-22). While many strategies are adopted for improving the growth of this sector, an important but often ignored factor is the environment, most particularly the air quality.

* Corresponding author: sidra.safdar@uvas.edu.pk
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The air contains high levels of dust, particulate matter and microorganisms inside the livestock facilities, which may cause respiratory diseases in the animals and even the workers at these farms (Eduard *et al*., 2009; May *et al*., 2012). Along with bioaerosols and particulate matter (PM), mixture of gases like methane (CH_4), ammonia (NH_3), hydrogen sulfide (H_2S), carbon dioxide (CO_2), and toxins are also reported in the air of livestock farms (Heederik *et al*., 2007; Guidry *et al*., 2017). It has been concluded from multiple studies that farm workers and even those residing in the vicinity of livestock farms are more at risk of contracting various respiratory diseases such as asthma and chronic obstructive pulmonary diseases (COPD) (Kauffmann *et al*., 2002; Smit *et al*., 2014; Borl´ee *et al*., 2015; Casey *et al*., 2015; Fontana *et al*., 2017; Douglas *et al*., 2018; Guillien *et al*., 2019; Schultz *et al*., 2019). Such situation makes the animals too more vulnerable to respiratory diseases along with their caretakers.

Air quality in Pakistan is already severely compromised with major cities topping the charts in terms of worst air quality in 2022 again. This situation combined with the annual smog episodes, particularly in Central Punjab region brings out dire consequences for the human beings while the effects on livestock also cannot be ignored since inhalation is a spontaneous process for all living beings. Recent researches have proved the relationship between poor air quality and respiratory diseases and increased mortalities in populations. In fact, in the year 2019, 99% of the global population was inhaling air that did not meet the air quality guidelines laid out by the World Health Organization. Moreover, ninety-one percent of premature deaths caused by poor air quality out of the 4.2 million deaths in 2016 occurred in low and middle income countries including South East Asia and the West-Pacific regions (Baccarelli *et al*., 2011; WHO Fact Sheet, 2022).

Among the common diseases related to bioaerosols in livestock farms, haemorrhagic septicaemia is one of the important diseases resulting in high mortality in cattle and buffalo in Asia (Benkirane and De Alwis, 2002). The causative agent of haemorrhagic septicaemia is *Pasteurella multocida* (Kuranasree, 2016). Likewise, bovine tuberculosis is the disease of dairy animals and hence of zoonotic importance, the causative agent of this disease is *Mycobacterium bovis* (Radostits *et al*., 2000). Still there are many more whose presence in the air can lead to a variety of diseases such as bovine pneumonia or bovine respiratory disease through air-borne transmission in livestock sector (Eames *et al*., 2009). As a matter of fact, bovine respiratory disease complex (BRDC) stays the most well-known and economically significant disease affecting cattle and buffaloes across the globe (Lubbers and Turnidge, 2015; Wolfger *et al*., 2015) and economic wastes to the United State feedlot industry have been reported as 1 billion dollars per year in labor costs, drug costs and death (Griffin *et al*., 2010). While clinical signs related with BRD show a fast onset, exact diagnosis of BRD shows a critical challenge (Wolfger *et al*., 2015) since it is caused by multiple agents. The viral respiratory diseases such as IBR, BVD, PI3, BSRV or bacterial species such as *Pasteurella haemolitica, Pasteurella multocida, Haeomophilus somnus, Staphylococcus aureus, Mycoplasma bovis* and *Clostridium perfringens* (Uzal *et al*., 2002; Fernandez-Miyakawa *et al*., 2007).

Despite many studies reporting the air quality in urban centers, livestock farms are not monitored regularly in this context. There are no studies on air pollution monitoring, particularly particulate matter from cattle farms from Pakistan and very few globally including those by Szulc *et al*. (2020) and Guo *et al*. (2022). However, such studies have been reported for poultry farms across the country as well as from different parts of the world (Yasmeen *et al*., 2020b). Livestock has been known to be a major contributor of greenhouse gas emissions including both methane and carbon dioxide (Hussain and Rehman, 2022). However particulate matter and heavy metals or potentially toxic trace elements (PTTEs) also need to be regularly monitored owing to their harmful impact. Moreover, composition of the air-borne microflora is also essential so as to prevent disease outbreaks and air borne transmission of diseases in livestock.

The current study is designed to investigate the air quality of large ruminant's farms in terms of its composition. The study will be useful in determining the overall environmental health status of animal farms and the factors responsible for spread of disease through air.

MATERIALS AND METHODS

Selection of study sites

Livestock farms were selected from two districts across Lahore Division i.e., Lahore and Kasur. The selection was made on the basis of information obtained from outdoor clinic of the University of Veterinary and Animal Sciences, Lahore (UVAS) regarding recent respiratory disease outbreaks (such as BRD, hemorrhagic septicemia) in different farms. The farms were visited and consent was obtained from the owners before final selection. The selected farms (Fig. 1; map) were divided into two groups:

Group A: Farms with no recent disease outbreak (in the last two years) (n = 5)

Group B: Farms with history of disease outbreak (in the last two years) (n = 5)

After selection, general information about the farm was obtained such as age of farm, number and type of animals, number of workers, hygiene maintenance, structure of farm and sheds, vaccination schedule, ventilation practices, disease history, presence of veterinary doctor or veterinary assistant.

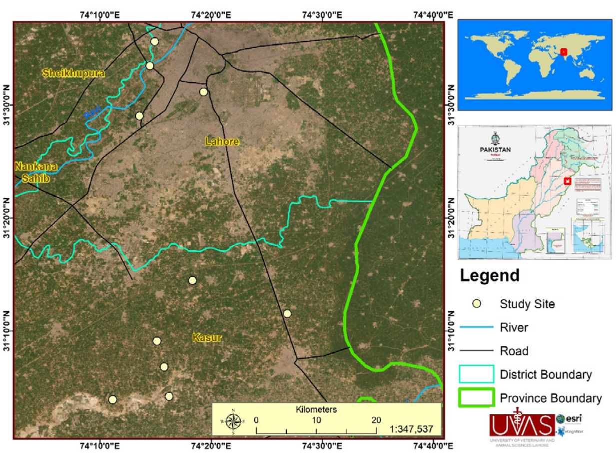

Fig. 1. Map of the study sites.

Spatio-temporal monitoring of air quality of animal farms

Monitoring is the foremost step in determining the status of healthy air in any microenvironment. For this purpose, air quality was monitored during the winter and summer season at the selected animal farms. The parameters monitored included temperature, relative humidity, fine and coarse particulate matter ($PM_{2.5}$ and PM_{10}), potentially toxic trace elements (PTTEs), carbon dioxide, and bioaerosols. The equipments for monitoring of PM were placed at a height of 1 meter above ground and away from any interfering sources such as in the walkways or near doors and windows and run for eight h at each site from 8:00 am to 4:00 pm during both seasons.

Particulate matter and potentially toxic trace elements (PTTEs)

Particulate matter was monitored using a real-time particulate counter, Gradko Laser Air particle counter DC-1700 which simultaneously counts $PM_{2.5}$ and PM_{10}. The data obtained was tabulated and values for $PM_{2.5}$ were converted from number count to micrograms per meter cube using the Dylos conversion formula (Arling *et al.*, 2010). This conversion formula has been compared and verified by Franken *et al.* (2019) to be a reliable tool for indicating the extent of pollution at the site. A volumetric air sampler (air flow rate of 20l/sec) fitted with pre-weighed Whatmann 41 filter paper was run in parallel for determining elemental composition of the suspended dust. After 30 min the filter was removed and kept in metal free containers till further assessment. The filter papers were digested to extract out the heavy metals including lead (Pb), Nickel (Ni) and Zinc (Zn) following wet digestion through HCl and HNO_3 in 2:1 ratio the digestate was diluted up to 25 ml with deionized water and subjected to analysis through Atomic Absorption Spectrophotometer (Hitachi Z-8230) for detection of trace metals including lead (Pb), nickel (Ni) and zinc (Zn). Carbon dioxide was measured by running a real time CO_2 sensor along with temperature and relative humidity.

Biological aerosols

Bioaerosols were sampled by using both passive (gravimetric sampling) and active sampling. For passive sampling, agar coated petri plates in triplicates were exposed for 30 min at each farm. After exposure the pre-labelled plates were sealed and stored at 4°C in an icebox till further analysis. For active sampling, a volumetric pump (air flow rate = 20l/sec) fitted with mixed cellulose ester membrane filter (47 mm diameter, pore size 0.22 µm) were run in parallel for 30 min. The filters were carefully removed and immediately transferred to falcon tubes containing phosphate buffer saline solution till further analysis.

For microbial analysis, the filter papers present in PBS solution were vortexed and serial dilution were prepared. 10^{-4} dilutions were prepared and 100 micro meter of dilution was plated on nutrient agar in duplicates. These and the other agar plates exposed for passive sampling were then incubated at 37°C for 24 h. After incubation the number of colonies on each plate was counted and colony forming units (CFU/cubic meter) calculated following the Omelyansky (1940).

N = 5a. 104 / (b.t)

Where N is colony forming units per m^3 (cfu/m^3); a is no. of colonies per petri dish, b is surface area of dish (cm^3), t is exposure time (min).

The samples were cultured and identified through colony morphology and biochemical testing following Bergey's Manual of Determinative Bacteriology (1994). Primary growth was examined for cultural and morphological characteristics, colony morphology was observed in terms of size, density and texture. For microscopic evaluation smear from different colonies was prepared followed by Gram staining and observed under oil emersion lens of microscope.

RESULTS AND DISCUSSION

Air quality is a major issue worldwide and while we focus more on human exposure, other living beings also are being equally exposed to same air we breathe. Livestock farming is a major component of agriculture sector and hence animal health has a significant impact on the economy. While there are a few studies which have reported particulate matter and bioaerosols from cattle farms around the world (Szulc *et al.*, 2020; Guo *et al.*, 2022), most of the studies worldwide are reported from poultry farms and swine farms; the latter not practiced in Pakistan. There are a few studies conducted on air quality assessment and its impacts in poultry farms of Pakistan (Yasmeen *et al.*, 2019, 2020a, b) while animal farms such as cattle, sheep and goat farms have not been reported once yet from the country. The current study is a first one to describe the air quality in cattle farms from Pakistan and was designed to evaluate the seasonal load of some major air quality parameters including particulate matter and bioaerosols at selected livestock farms of Lahore division. The farms with no disease outbreak in the recent two years (Group-A) were generally old as compared to farms with disease history in the last two years (Group-B). Moreover, three farms in group-B had no full-time veterinary assistant and the animals were vaccinated after mortalities due to disease spread. Cleaning schedule also varied greatly between both categories with group-A farms cleaned more frequently (Table I).

Table I. Description of selected animal farms for air quality monitoring.

Group	No. of animals	Farm area	Cleaning	Farm age (years)	Workers (n)	Surroundings	Feeding	Solid waste disposal	Water waste	VA
A1	170	5 Acre 7 Shed	After 2 h	5	25	Near highway	Silage +Wanda	Within shed arranged	Open drainage	Available
A2	180	2 Acre 3 Shed	Twice daily	4	13	Near motorway	Silage +Wanda	Within shed arranged	Open drainage	Available
A3	120	1 Acre 2 Shed	Twice daily	3	11	Agricultural	Silage +Wanda	Within shed arranged	Open drainage	Available
A4	200	2 Acre 4 Shed	Thrice daily	7	18	Agricultural	Silage +Wanda	Within shed	Open drainage	Available
A5	260	4.5Acres 3 Shed	Twice daily	4	22	Agricultural	Silage +Wanda	Within shed	Open drainage	Available
B1	275	1 Acres 2 Shed	Once daily	1.5	30	Near highway	Silage	Open	With shed	NA
B2	120	0.5 Acre 1 Shed	Once daily	2	12	With highway	Silage	Open	With shed	NA
B3	90	1 Acre 14 Shed	Once daily	3	05	Agricultural	Wanda	Open	With shed	NA
B4	180	3 Acres 3 Shed	Once daily	4	25	Agricultural	Silage	Open	With shed	Available
B5	155	2 Acres 2 Shed	Once daily	2	20	Agricultural	Silage	Open	With shed	Available

VA, veterinary assistant; NA, not available.

Table II. Air quality parameters monitored in selected livestock farms.

Group	$PM_{2.5}$ (µg/m³)	PM_{10} (Particle count #)	Temp. (°C)	Humidity (%age)	CO_2 (ppm)
A	3.29 ± 1.82	14.8 ± 11.52	26.56 ± 15.55	56.58 ± 22.95	405.28 ± 73.09
B	3.81 ± 3.67	16.07 ± 11.63	26.56 ± 14.85	59.1 ± 45.41	492.11 ± 77.56
P-value	0.013*	0.133	0.966	0.335	0.029*

Data are Mean ± SD, Independent sample t test, level of significance 5%, *significant difference.

Table III. Count (Mean±SD) of coarse particulate matter (PM_{10}) at the selected farms during summer season (S) and winter season (W).

	Summer					Winter				
	1	2	3	4	5	1	2	3	4	5
A	21.23±13.71 (3-76)	21.42±14.55 (3-66)	21.57± 10.76 (3-50)	21.34±10.49 (3-45)	22.13±10.47 (3-44)	7.15±4.79 (1-21)	7.68±5.63 (1-25)	9.63±6.73 (1-33)	7.21±4.90 (1-21)	8.63±5.18 (1-20)
B	22.52±10.65 (3-44)	21.89±9.98 (3-44)	21.55±9.23 (6-44)	22.47±11.54 (3-55)	22.76±15.61 (6-95)	8.73±5.61 (1-22)	8.63±5.14 (1-21)	10.10±8.99 (1-55)	9.55±5.95 (1-25)	12.42±11.21 (1-45)

Air quality levels in sampling sites

Particulate matter was monitored through two seasons and the mean concentration of $PM_{2.5}$ was recorded to be 3.29±1.82 µg/m³ in group-A farms and 3.81±3.67 µg/m³ in group-B farms (Table II). While the means levels do not exceed the national environmental quality standards (NEQS), the seasonal impact and variations during different activities need to be considered (Table IV and V; Fig. 2). Outputs of one-way ANOVA indicated a significant difference in $PM_{2.5}$ concentrations in the air of both groups of farms during the summer season (p = 0.006) while the difference was not pronounced during the winter season (p = 0.575). A study from pig farms in China reported higher levels of fine particulate matter in the range of 60 µg/m³ to 200 µg/m³ (Shang *et al*., 2020).

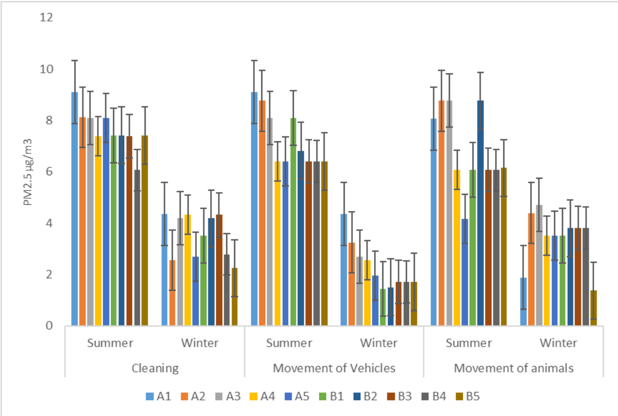

Fig. 2. Mean concentration of $PM_{2.5}$ µg/m³ during different activities carried out the selected farms (Cleaning: floor sweeping; movement of vehicles: movement of large vehicles including tractor trollies for bringing in fodder or transport of animals; Movement of animals: animals being moved around the farm or for grazing in the surrounding fields).

Paired t test was applied on the data to observe any significant seasonal impact upon PM load. There was a statistically significant decrease in $PM_{2.5}$ concentrations from summer season (M = 4.84, SD = 3.62) to winter season (M = 2.33, SD = 0.97), t (370) = 13/328, $p < 0.0005$. The mean decrease was 2.51 with a 95% confidence interval ranging from 2.14 to 2.88. An increase of $PM_{2.5}$ levels in summer season of livestock farms (Pig farms) has been reported by Tang *et al*. (2020), However in contrast to the results obtained, the PM levels in other ambient and indoor environments have been reported by many researchers to be higher during the winter season as compared to the summer season (Sidra *et al*., 2015). Number count of PM_{10} was also higher in group B farms in comparison with group A farms (Tables II and III).

Livestock is a major contributor towards greenhouse gas (GHG) emissions including CO_2 and Methane. A recent study found out that CO_2 production is significantly associated with the stock of livestock in Pakistan and a single percent increase in this overall stock would result in 0.45% increase in CO_2 emissions (Ullah *et al*., 2018). In our study the mean CO_2 levels at the selected farms were higher in group B farms as compared to group A farms, the standard deviation also indicated larger deviation of data in group B farms (Tables II, IV and V). Moreover, it was found that CO_2 levels were higher in group B farms during the winter season (601.11 ppm in contrast to 383.12 ppm in summers) (Table IV). The major reason attributed to this increase was the burning of wood and coal by the workers to keep themselves warm during the cold season.

Apart from seasonal influx, daily routine activities also contribute towards the levels of fine particulates in any microenvironment. In our case the major activities were identified to include cleaning of the farms, movement of vehicles such as tractors around the farm area and most importantly movement of the animals. The time of the day also had a major role in defining the concentration of PM as indicated by the outputs of one-way ANOVA (p = 0.00) with mornings being busier and afternoon generally more peaceful (Table V). Cleaning was the bigger contributor towards $PM_{2.5}$ levels as evident in Figure 2.

Table IV. Seasonal Variations of air quality parameters monitored in selected livestock farms.

Group	Seasons	$PM_{2.5}$ (µg/m³)	PM_{10} (Particle count)	Temperature (°C)	Humidity (% age)	CO_2 (ppm)
A	Summer	5.28 ± 4.67*	21.54 ± 12.01*	41.39 ± 4.21	38.69 ± 10.96	376.82 ± 63.48
	Winter	2.34 ± 0.96*	8.06 ± 5.52*	11.72 ± 4.98	74.47 ± 17.1	433.75 ± 71.08
B	Summer	4.28 ± 1.94*	22.24 ± 11.57*	40.81 ± 3.98	41.59 ± 19.53	383.12 ± 96.15
	Winter	2.3 ± 0.9*	9.89 ± 7.79*	12.32 ± 4.28	76.61 ± 56.01	601.11 ± 75.88

Data are Mean ± SD, Independent sample t test, level of significance 5%, *significant difference

Table V. Comparison between different durations of times for air quality monitoring in the selected animal farms.

Group	Time	$PM_{2.5}$ (µg/m³)	PM_{10} (Particle count)	Temperature (°C)	Humidity (% age)	CO_2 (ppm)
A	Morning	$4.79^a \pm 5.41$	$19.64^a \pm 13.39$	$22.01^c \pm 14.68$	$67.4^a \pm 20.32$	$433.95^a \pm 89.31$
	Noon	$4.16^a \pm 2.42$	$14.55^b \pm 11.11$	$26.5^b \pm 16.79$	$59.52^b \pm 24.95$	$385.75^b \pm 56.27$
	After-Noon	$2.37^b \pm 1.28$	$9.83^c \pm 6.6$	$31.55^a \pm 13.54$	$41.68^c \pm 13.9$	$395.39^b \pm 59.33$
B	Morning	$3.73^a \pm 1.69$	$20.29^a \pm 13.24$	$22.49^c \pm 13.78$	$66.77^a \pm 20.4$	564.15 ± .28
	Noon	$3.69^a \pm 2$	$16.18^b \pm 11.68$	$26.61^b \pm 15.92$	$59.46^{ab} \pm 23.95$	386.4 ± 90.46
	After-Noon	$2.37^b \pm 1.37$	$11.37^c \pm 7.15$	$30.93^a \pm 13.59$	$50.4^b \pm 73.17$	528.6 ± 92.78

Data are Mean ± SD, One-way ANOVA, level of significance 5%, *significant difference.

PTEs in ambient air

The air-borne levels of PTEs were measured in animal housing facilities. Three trace metals were analyzed i.e., lead (Pb), nickel (Ni) and zinc (Zn). The trend of occurrence of trace metals were observed as Pb >Zn > Ni (Fig. 3). The levels of all metals were however within the limits of NEQS.

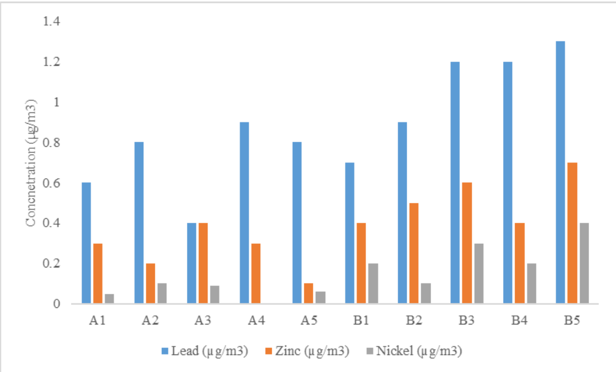

Fig. 3. Air-borne levels of PTEs [Lead (Pb), Nickel (Ni) and Zinc (Zn)] in healthy and diseased farms.

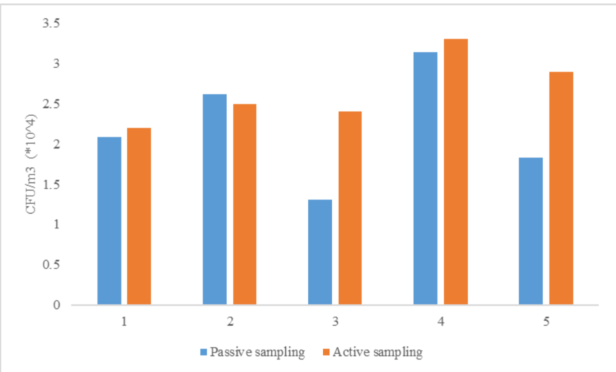

Fig. 4. Comparison of bacterial colonies present in air of diseased livestock farms.

Air-borne microflora at selected farms

In livestock housing facilities, airborne microorganisms are present in a huge quantity that can affect animals and workers (Riccardi *et al*., 2021). Samples obtained from passive sampling contain a large number of microorganisms. Major species identified included *Staphylococcus aureus* and *Pasteurella multocida* and confirmed from sanger sequencing). Overall, the colony forming units at the respective farms through passive sampling were 2.09×10^4, 2.62×10^4, 1.31×10^4, 3.14×10^4 and 1.83×10^4 CFU/m^3 while the results of volumetric/active sampling indicated colony forming units to be 2.20×10^4, 2.50×10^4, 2.40×10^4, 3.30×10^4 and 2.90×10^4 CFU/m^3 (Fig. 4). Bacterial colonies which were identified under the microscope were round-shaped as gram-positive bacteria identified as *Staphylococcus aureus* (Fig. 4).

Assessment of aero-biome of the selected livestock farms sheds some light on the overall housing conditions and contaminant levels in their air. While the overall levels may not be higher than the prescribed limits, it is important to estimate the ecological footprint of livestock farming and for this purpose the atmosphere is also equally important. As per our observations, while the PM levels were within the WHO guidelines, farms where cleaning was not frequent or without proper drainage, there was an increase in PM levels. Similarly, the CO_2 levels also need to be considered as livestock sector is already a major contributor of GHG emissions. Proper ventilation, regular cleaning and maintain proper drainage can not only improve the general environment but the well-being of the animals as well. Aerobiological screening also indicated that timely vaccination of animals along with proper cleaning and proper drainage of the waste had a significant impact upon the bacterial load detected in air.

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IRB approval

The study was approvedby University of Veterinary and Animal Sciences, Lahore, Pakistan through letter no. DAS/418 dated 28-01-2022.

Ethics statement

All research was conducted in accordance with ethical principles.

Statement of conflict of interest

The authors have declared no conflict of interest.

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